CHAPTER FIVE

"It is not the strongest of species that survive, nor the most intelligent, but the ones most responsive to change."

Charles Darwin (1861) *On the Origin of Species by Means of Natural Selection*, 3rd ed. John Murray, London

5 Variation in Morphology, Productivity and Dispersal in Selected Species

In Chapter Four only 25% of the variance in species distribution was explained by the physical and chemical factors measured on the shale bings. Significant groups of species were revealed to be associated with gradients in the physical and chemical environment and these species were shown to be associated with distinct mechanisms of dispersal or growth habit. The aims of this part of the investigation were to explain at least some of the remaining variance by studying the patterns of distribution and growth in a few selected species in relation to seed dynamics: measured as availability, dispersability and fecundity, and morphological plasticity represented by plant height. The questions being addressed are:

How great a determining factor in the establishment of vegetation is the availability of seeds and spores in seed rain and the seedbank?

Is phenotypic plasticity a major contributor to vegetation dynamics at individual species level?

Is any variation in seed production within a species correlated with either height or number of flower-heads?

Will local site conditions have a direct effect on the size of individual plants within a species, measured as height above ground, and hence on the number of flower-heads and seeds produced?

In previous chapters species associations, general vegetation patterns and correlations have been identified using all species, or the most frequent species, recorded on all sites. As suggested in Chapter Four it would be impractical to gather the additional information required from all 211 species recorded on the shale bings,

or even to use the 100 most frequent species. In this chapter within species variation in morphology and the mechanisms of seed dynamics are investigated using the nine species that were identified in the previous chapter as being representative of some feature of vegetation distribution in relation to the physical and chemical environmental variables measured in the bing habitat.

5.1 Plasticity and seed ecology

Phenotypic variation can often be more clearly perceptible than genotypic variation within the same species. Plasticity enables individuals of a species to survive in non-optimal conditions, making it possible for the species to survive over several generations, until new, better-suited, genotypes are produced. The inherent plasticity in most plant species enables individuals, developing from the seedlings of a single plant, to vary considerably in height, number of flowering heads produced and number of seeds per flowering head.

The availability of seeds and other propagules in a primary succession is a major contributing factor to the patterns of colonisation. The input of propagules is not limited to those produced by populations of species in the immediate locality. Initial recruitment is often from long range dispersal, particularly when the new substrate covers a large area or is very high compared to the surrounding landscape (Cronk and Fuller 1995), as is the case on shale bings. As establishment of species increases there are additional sources of invasion within the site, from the seedbank, and from seed rain. Legg *et al*. (1992) related the numbers of seeds produced by *Calluna vulgaris* to those recorded in seed rain, the seedbank and to the final outcome of the vegetation. Seed rain patterns in New Zealand were linked to vegetation patterns in successional seres by Dungan *et al*. (2001). Similar correlations are expected between the patterns of seed rain falling on the bings and the resulting vegetation.

The number of flowering heads and seeds per flowering head that are produced by individuals from the selected species is expected to be linked with variation in the height of adult individuals within the species recorded on the bing sites. Zammit and Zedler (1993) established that in the obligate-seeding shrub *Ceanothus greggii* height

was the major determinant of seed production and the total display area of flowers was shown to be relative to overall plant size in a study by Fabbro and Korner (2004). The viability of any seeds produced is expected to be equally important to the establishment of species in the vegetation succession. It would therefore seem appropriate to concentrate on these areas for further experiment and analysis.

5.2 Nine species

In many studies detailed examination is made of one dominant species (e.g. *Calluna vulgaris*, Legg *et al*., 1992) or species representing a particular ecological type (e.g. nitrogen fixing plants, Burden, 1980a: 1980b). The small-scale heterogeneity of vegetation recorded on the bing sites (up to 29 species in a single 2 m x 2 m quadrat) made selection of species on the basis of dominance alone very difficult. Recognised ecological types and functional groups of species (as described in Burden and Harper, 1980) were not associating as expected in the vegetation of the bings: partly because of the influence of planting and seeding and partly because of the early stages of development of vegetation types, as was demonstrated by the apparently wide range of National Vegetation Classification types identified in Chapter Three (3.1.3). Analysis of the baseline survey data from eight oil-shale bings did however identify groups of species that were representative of three local ecological habits:

1. Species belonging to the same family and sharing many ecological traits, that were rarely, if ever, recorded together on any of the bing sites despite being locally abundant (in West Lothian).
2. Species that were commonly recorded on the bings but that were rarely found in the surrounding environment.
3. Species that were abundant and common to both the bing sites and the surrounding habitats.

A further group representing species that were commonly recorded in the surrounding environment but that were rarely or never recorded on the bings could also have been investigated. Such a study was beyond the resources of this exercise and would have required the collection and analysis of large amounts of new data from a variety of sites in the surrounding countryside.

Nine 'weed' species (Table 5.1) that demonstrated these three ecological types were selected for further study and measurement as described in Chapter Four (Section 4.6.7). In this way the processes and mechanisms of vegetation succession could be described in terms of individuals within individual species that are representative of each of the described types.

Table 5.1 The Nine Species

The binomial, authority, common name(s) and family for each of the nine species used in the trials and data analyses in this chapter

Species	Authority	Common name	Family
Medicago lupulina	L.	black medick	Fabaceae
Trifolium campestre	Schreb.	hop trefoil	Fabaceae
Leucanthemum vulgare	Lam.	ox-eye daisy, moon daisy	Asteraceae
Tripleurospermum inodoratum	(L.) Sch. Bip.	scentless mayweed	Asteraceae
Reseda luteola	L.	dyer's rocket, weld	Resedaceae
Senecio viscosus	L.	sticky groundsel	Asteraceae
Holcus lanatus	L.	Yorkshire fog	Poaceae
Centaurea nigra	L.	common knapweed, hardheads	Asteraceae
Plantago lanceolata	L.	ribwort plantain	Plantaginaceae

Two pairs of species were selected to represent the first ecological type. The two species *Medicago lupulina* and *Trifolium campestre* represent one plant family, Fabaceae. The two species are similar in appearance (Figure 5.1) and share many ecological traits. Although they were recorded together on only one bing site, Addiewell south, the two species were distributed differently (*T. campestre* at the top and middle and *M. lupulina* in the excavated area). The two species were not recorded together on any of the other bings in the baseline survey, despite *T. campestre* appearing in 46 quadrats and *M. lupulina* in 40 (out of 340 quadrats). Species distribution maps of West Lothian demonstrate that variation of distribution between the two species extends throughout the county (Figure 5.1).

Leucanthemum vulgare and *Tripleurospermum inodorum*, Asteraceae family, are a second pair of similar looking species that were frequently recorded (Figure 5.2). *L. vulgare* was recorded in 124 quadrats on six sites, *T. inodorum* 38 quadrats on four sites. The two species were found on the same sites, and positions within sites,

but were only recorded growing together within nine quadrats at Clapperton on ground recently disturbed by planting.

Reseda luteola and *Senecio viscosus* were selected to represent the second ecological type. These species are not found locally in other habitats and were only recorded in areas of the bing sites with little or no competition from other vegetation (Figure 5.3). They were frequently recorded with *Myosotis arvensis* in quadrats with a large percentage of bare substrate, usually at high altitudes and on steeper slopes and have traits that are traditionally associated with early colonisers.

The final ecological type is represented by three species that are common perennial 'weeds' and are found over a wide range of habitats. *Holcus lanatus*, *Centaurea nigra* and *Plantago lanceolata*, were selected because they were constant to all bing sites, altitudes and aspects and were also commonly found in the surrounding habitats (Figure 5.4).

The nine species are all native, locally common weeds (Smith *et al*., 2002) and have been recorded consistently in numerous studies of colonisation over the last 100 years indicating that as well as being common weed species they are an important component of early successional vegetation. Brenchley & Adam (1915) recorded their appearance and fluctuations in abundance over more than 20 years in a study of two sites left fallow after cultivation. They were also recorded in vegetation surveys of a range of post-industrial sites including stone quarries (Hepburn, 1955; Davis, 1982) and disused pits (Hall, 1957). The germination characteristics of the nine species have been described in a major study of the Sheffield flora by Grime *et al*. (1981).

The nine species are all indicative of low shade, moist free-draining soils, neutral-basic soils and sites of intermediate fertility (Hill *et al*., 1999) and with the exception of *Centaurea nigra*, rely on a persistent seedbank as a major regeneration strategy (Hodgson *et al*., 1995). The autoecological traits, characteristics of reproduction and seed ecology for each species, as presented in these publications (Table 5.2), can be used for comparison with the measured data from this study.

Figure 5.1 ***Medicago lupulina*** **and** ***Trifolium campestre*** **(Fabaceae)**

Medicago lupulina *and* Trifolium campestre*, the two Fabaceae species (photographs from Bioimages, undated), demonstrating their similarity in appearance and growth form are displayed above distribution maps of both species adapted from Plant Life of Edinburgh and the Lothians (Smith* et al.*, 2002). Within West Lothian (the political county boundary is marked in bold where it does not follow the vice county boundaries)* M. lupina *and* T. campestre *were each recorded in 26 tetrads*[1] *and were recorded together in only 15 of these. There are a total of 134 tetrads within the political boundary of West Lothian (as described in Chapter Two), 54 of these are in (or partly in) Vice County 83 (Edinburghshire / Mid Lothian) and 80 are in Vice County 84 (Linlithgowshire / West Lothian).*

[1] "Each tetrad is made up of four 1 km x 1 km squares. One square in each tetrad was then chosen at random as the square to be surveyed." Plant Life of Edinburgh and the Lothians, page 184 (Smith *et al.*, 2002).

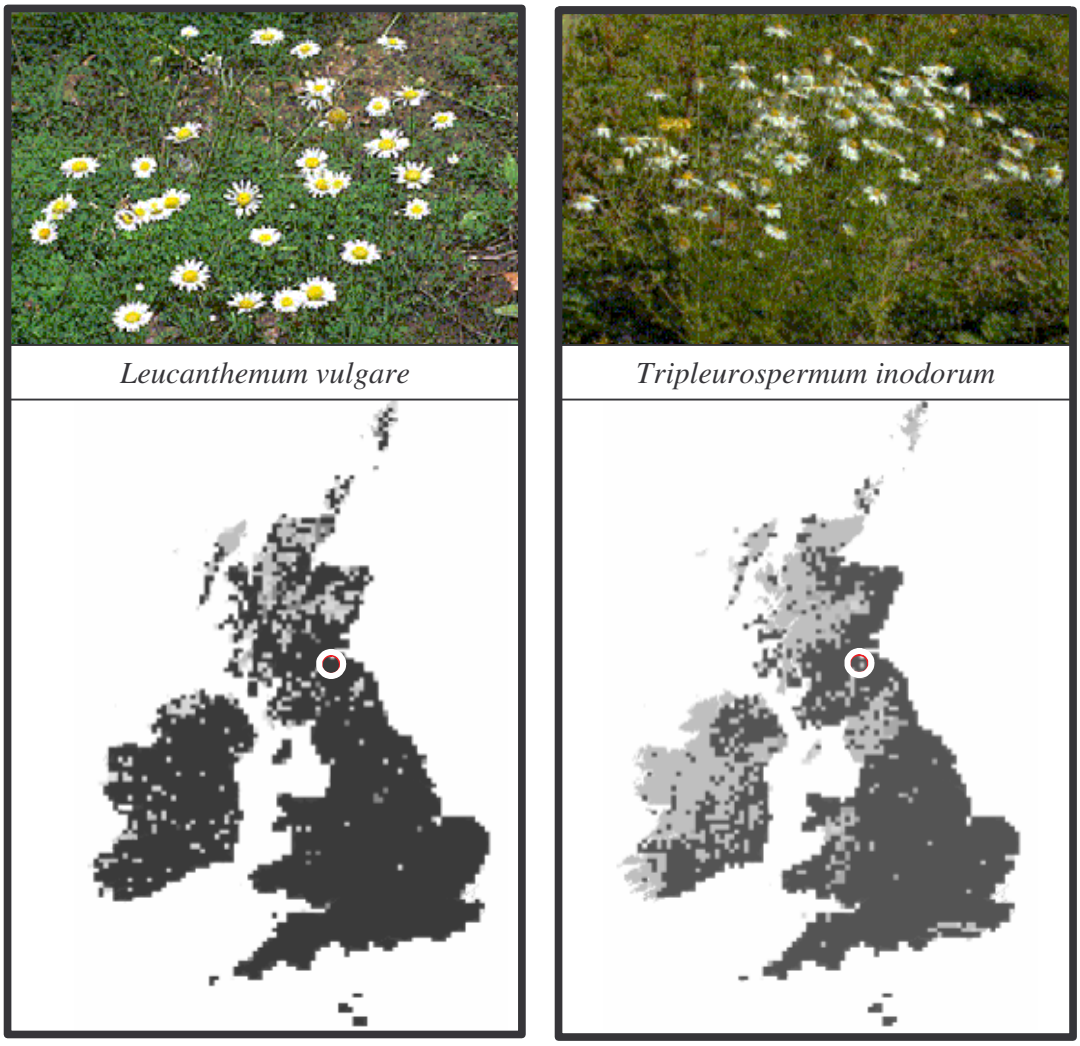

Figure 5.2 *Leucanthemum vulgare* and *Tripleurospermum inodorum* (Asteraceae)

Leucanthemum vulgare *and* Tripleurospermum inodorum *(photographs from Bioimages, undated), the two Asteraceae species, demonstrating their similarity in appearance, but different growth form are displayed with distribution maps of both species adapted from New Atlas of the British and Irish Flora (Preston* et al.*, 2002) showing the extensive distribution of these two species. White circles outline West Lothian and highlight that* T. inodorum *is less widely recorded in the county than* L. vulgare.

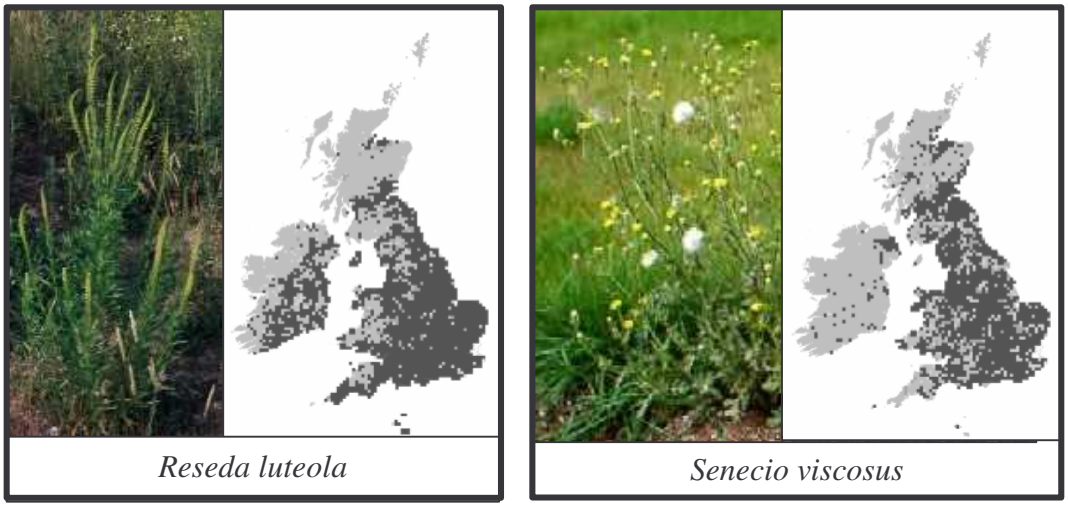

Figure 5.3 *Reseda luteola* and *Senecio viscosus*, the two 'bing species'

Reseda luteola *and* Senecio viscosus *(photographs from Bioimages, undated), two species that are commonly recorded on the bings but are rarely found in surrounding landscape. Distribution maps of the species adapted from New Atlas of the British and Irish Flora (Preston* et al.*, 2002) show that both species are sparsely recorded in much of Scotland.*

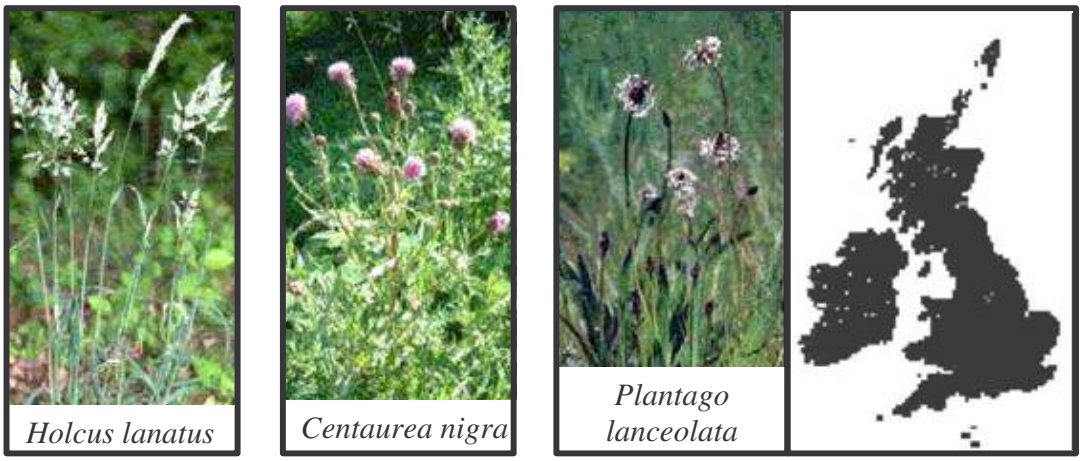

Figure 5.4 *Holcus lanatus*, *Centaurea nigra* and *Plantago lanceolata*, the three 'common species'

Holcus lanatus, Centaurea nigra *and* Plantago lancolata *(photographs from Bioimages, undated), three species that are commonly recorded on the bings and are very common in the surrounding landscape. The distribution map of* P. lanceolata *adapted from New Atlas of the British and Irish Flora (Preston* et al.*, 2002) shows the widespread distribution of this species throughout Britain. The distributions of* C. nigra *and* H. lanatus *are equally widespread.*

Table 5.2 The nine species their main autoecological traits, reproduction and seed characteristics

The nine plant species selected for detailed investigation are grouped as described in Section 5.2 above. Nomenclature follows Stace (1997) and the species are listed with information on life history, establishment strategy, regenerative status, seedbank, most common terminal habitat, soil pH, agency of dispersal and seed (dispersule) weight (Hodgson et al.*, 1995) and their allocated Ellenberg indicator values for light (L), moisture (F), reaction (R) and nitrogen (N) (Hill* et al.*, 1999).*

Habitat key: OUTCRP = rock outcrop; QRYl = limestone quarry spoil; ARABLE =arable; COAL = coal-mine spoil; CINDER = cinder tips; PASTe = enclosed pasture; WASTEl = wasteland on limestone strata; MEADOW = meadows.

Soil pH key: The numeral indicates the modal pH class for the species followed by a letter indicating the number of pH classes in which the frequency of the species exceeds 50% of that in its modal class. Thus 5a indicates that the species is most frequent within the range 5-5.9 and has a range of only 1pH unit. In contrast 4d indicates a mode within the interval pH 4-4.9 and a range of 4 pH units.

Agency of dispersal key: ANIMa = animal dispersed, dispersule with awn; ANIMm = animal dispersed, dispersule adhesive from mucilage; UNSP = unspecialised; UNSPag = unspecialised but dispersed as a result of agricultural practices; WINDc = wind dispersed, seeds small and shed from a capsule held above the surrounding vegetation; WINDp = wind dispersed, dispersule plumed or wrapped in hairs.

Seed weight key: 1 ≤ 0.20mg; 2 = 0.21-0.50mg; 3 = 0.51-1.00mg; 4 = 1.01-2.00mg; 5 = 2.01-10.00mg; 6 > 10.00mg.

Life history key: Aws = annual; As = summer annual; Aw = winter annual; P = perennial; B = biennial.

Established strategy key: primary strategies are C = competitor; S = stress-tolerant; R = ruderal; secondary strategies are CR = competitive ruderal; SC = stress-tolerant competitor; SR = stress-tolerant ruderal; CSR = CSR strategist; there are twelve further strategy intermediates between these seven (e.g. CR/CSR) making a total of nineteen types.

Regenerative status key: Bs = a persistent bank of buried seeds or spores; S = seasonal regeneration by seed; V = lateral vegetative spread; W = regeneration involving numerous widely dispersed seeds or spores.

Seedbank key: 1=transient: seed rarely persisting for more than 1 year; 2 = short-term persistent: seed persisting for more than one year but usually less than five; 3 = long-term persistent: seeds persisting for at least five years and often much longer

Indicator values key: L (light) ranging from 1 (plant in deep shade - none in UK) to 9 (plant in full light); F (moisture) ranging from 1 (indicator of extreme dryness) to 12 (submerged plant); R (reaction) measured as "the gradient of soil acidity and lime content"[2] ranging from 1 (indicator of extreme acidity) to 9 (indicator of base reaction); N (nitrogen) is a general indicator of soil fertility ranging from 1 (extremely infertile) to 9 (extremely rich).

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[2] The reaction scale is not synonymous with the divisions of the full pH scale (0-14) but is a scale indicative of the range of acidity and alkalinity found in European soils (Ellenberg, 1988).

Species	Habitat	Soil pH	Agency of dispersal	Seed weight	Life history	Established strategy	Regenerative status	Seed bank	Indicator values			
									L	F	R	N
Medicago lupulina	OUTCROP	7a	UNSP	5	A/P	R/SR	Bs	3	7	4	8	4
Trifolium campestre	OUTCROP	6c	ANIMa	2	Aw	SR	S,Bs	3	8	4	6	4
Leucanthemum vulgare	QRYl	7a	UNSPag	2	P	C/CSR	V,S,Bs	3	8	4	7	4
Tripleurospermum inodorum	ARABLE	6c	UNSPag	2	Aws	R	S,Bs	3	8	5	6	6
Reseda luteola	COAL	6b	WINDc	2	B/P	R/CSR	Bs	3	7	4	8	6
Senecio viscosus	CINDER	7c	WINDp	3	As	R	W,Bs	3	8	5	7	6
Holcus lanatus	PASTe	5c	UNSP	2	P	CSR	V,S,Bs	3	7	6	6	5
Centaurea nigra	WASTEl	7c	UNSP	5	P	CSR	V,S	2	7	5	6	5
Plantago lanceolata	MEADOW	7c	ANIMm	4	P	CSR	V,Bs	3	7	5	6	4

The baseline survey established that all nine of the selected group were within the top fifty species (out of 211) by both frequency and accumulated percentage cover on the bing sites. Four of the species selected were among the twenty most frequently recorded and among the twenty species with the highest accumulated percentage cover (*H. lanatus*, *L. vulgare*, *P. lanceolata*, and *C. nigra*). *S. viscosus* was also in the twenty most frequently recorded species and *M. lupulina* in the twenty species with highest accumulated percentage cover. The species were all recorded on the three main positions-on-site, top, middle and base, although *T. inodorum* and *M. lupulina* were not recorded on the plateaux and *C. nigra*, *R. luteola* and *L. vulgare* were not recorded on the excavated sites. The nine species grew successfully at all aspects (north, south, east, west, northeast, southeast, southwest, northwest) and on slopes up to 60 degrees from horizontal except *T. inodorum* which was only recorded on slopes up to 40 degrees. *S. viscosus* and *R. luteola* were not recorded in quadrats with more than 40% ground cover although the other seven species were recorded in quadrats containing all levels of ground cover. This suggested that variation in physical environmental gradients could be ruled out as a major determining factor in the distribution of the species with the possible exceptions of *T. inodorum* (angle of slope) and *S. viscosus* and *R. luteola* (competition from other species). These three species were also absent from quadrats recorded on the old managed areas of the bings as defined in Chapter Four (Section 4.2.1).

5.3 Measurements and trials

The extent of morphological variation within and between species was measured and counted in the nine selected species: height (representing plasticity), number of flowering heads produced and number of seeds produced per flowering head. A series of field experiments and glasshouse trials were established to measure the species components of the substrate seedbank and seed rain on the bing sites, and the germination potential of seeds collected in the field. The resulting data were analysed to establish whether height varied within and between species as a response to individual environmental factors and to determine any relationships between variability in height and the mechanisms and processes of seed production, dispersal

and germinability within each species. The resulting information demonstrated to what extent the variation in seed dynamics and morphological variability within these species is the cause of variance in the distribution of vegetation.

5.3.1 Plant height and seed production

The number of individual plants growing in each 16 m^2 quadrat block (as described in Chapter Four) was recorded for each of the nine species. The heights of ten randomly selected plants of each species were measured (from the surface of the substrate to the highest point of the plant) and the total number of flowering heads (immature and mature) per plant were counted and recorded. Ten representative, mature flowering heads were selected from each plant and the number of seeds were counted and recorded. If ten individuals of any species were not represented in the quadrat and plants were growing within a further 1 m of the block in any direction then these were measured but not included in the plant count for the quadrat. If there were no flowering heads on the selected plant or no seeds in the selected flowering head these were recorded as zero values. Forty quadrat blocks were surveyed in this way, 12 at Oakbank, 12 at Clapperton and 16 at Greendykes. Counting was abandoned at Mid Breich due to overlapping ranges at the top and middle positions-on-site on this very small bing. All further trials and measurements relate to only three sites: Greendykes, Oakbank and Clapperton.

5.3.2 Seed and seedling identification

Samples of seed were collected from as many species as possible during the field seasons, while still attached to the plant, and were used to produce a seed 'herbarium' for use in the identification of samples from seed rain and the seedbank. An herbarium of seedling stages was compiled for grasses, and other vascular species that were expected to have less easily identifiable leaf morphologies, by germinating known seeds and harvesting samples at regular intervals.

5.3.3 Glasshouse germination trials

Seeds for germination trials were collected in late autumn 1999 from plants of each of the nine species that were growing in and around the fixed quadrat blocks. Two hundred and forty seeds from each species were sown in 12 individual plant pots

(10 cm diameter) filled with vermiculite growing medium, twenty seeds to a pot. Thirty six pots were prepared without seeds as controls. Nine pots, one sown with seed from each of the species, and three control pots were randomly positioned on each of 12 trays (a total of 144 pots). The trays and their contents were then randomly positioned in the heated mist unit of a glasshouse on 10th October 1999.

The trays were first inspected after one week, then on Monday, Wednesday and Friday of each week thereafter. Germinated seeds were counted and removed with as little disturbance as possible on each occasion. After 8 weeks, when there had been no new germination recorded for 2 weeks, the trays were removed to cold frames and monitored weekly. There was then no germination for several months until a series of frosts at the end of April 2000 after which additional germination was recorded. Weekly monitoring was terminated in August 2000 after three months with no further germination from any of the species.

5.3.4 Soil seedbank trial

A seedbank trial was set up to examine the availability of propagules from this source. Substrate samples for seedbank analysis were collected from the same fixed quadrat blocks as were used for chemical analysis (Chapter Four, section 4.2.2) plant measurements (5.3.1) and collecting seeds (5.3.2). The 300 g that was retained from each of the bulked and concentrated substrate samples (Chapter Four, Section 4.2) was spread in a 25 mm layer (Fenner, 1985) over sharp sand in a 150 mm x 200 mm seed tray. The resulting 100 seed trays, containing 50 surface soil samples and 50 unweathered substrate samples, plus an additional ten control trays containing only sand were set out randomly in the mist unit of a heated glasshouse. The trays were monitored for 3 months (Thomson *et al*., 1997) and seedlings were identified by comparison with a collection of seedlings from known parent plants (5.3.2) and counted as soon as possible after they emerged, then removed. Any unrecognised seedlings were identified using Williams & Morrison (1987) and Hanf (1974). All individuals, of all species germinating, were recorded. At the end of the 3-month period the substrate in each tray was hand sorted to find any propagules that had not germinated.

5.3.5 Seed traps

The open nature of the shale bings, both to the elements and the public, meant that standard (commercial) equipment for seed trapping could not be used. Seed traps were designed to be as inconspicuous as possible. The traps were constructed from empty 2 litre soft drinks bottles, wire mesh (5 mm x 5 mm to avoid trapping small mammals) and either sections of 20 denier tights or filter paper (Figure 5.5a). Prototypes of both versions were tested in various positions in a garden to ensure that they functioned effectively. The two versions of the trap worked equally well and I elected to use the filter paper version because it was slightly easier to construct.

The seed traps were set at ground level (Figure 5.5b), one in each of the forty quadrat blocks used for substrate analysis. The traps were placed in a randomly selected hole, one of ten, left by the removal of substrate for the seedbank trial.

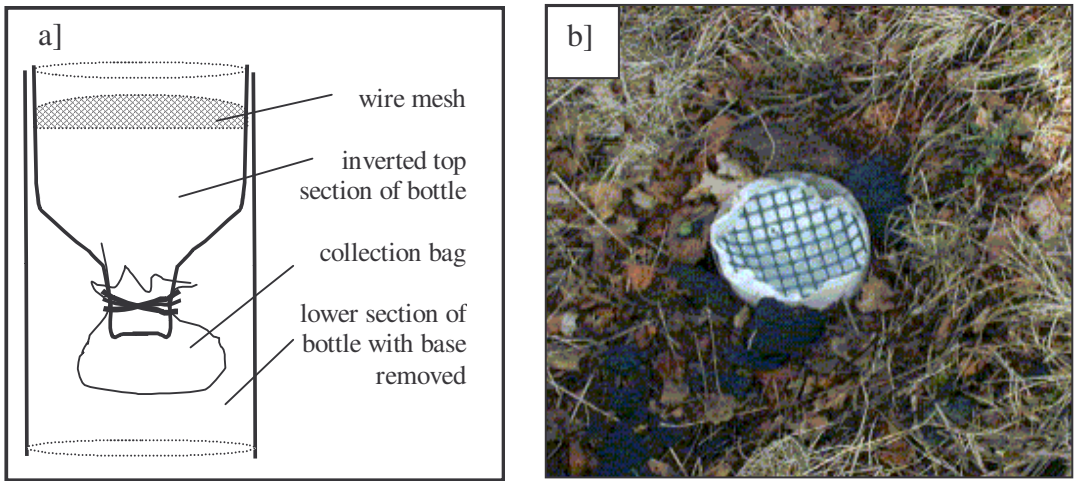

Figure 5.5 Seed traps

a] Seed trap with collection bag made from sections of tights fixed with an elastic band and b] photograph of a seed trap with filter paper insert in situ *at Oakbank bing.*

Unfortunately there were no results from the seed traps on any of the sites. Both variations of the seed trap were set out on two or more occasions but when revisited for seed collection were found to have been scattered and destroyed by Corvids (probably rooks or jackdaws) at all sites and this attempt to measure seed rain had to be abandoned.

5.3.6 Analysis of data

The correlation analysis tool in Microsoft Excel was used to measure any relationship between the data sets of morphological measurements for each of the nine species; plant height, population density, number of flower-heads and number of seeds. The coefficient of variance (CV) was calculated to compare within species variation between the morphologically different species (CV = 100 x SD / mean).

Variation between the seedbank data for the nine species linked to either the surface samples or the unweathered samples was tested by calculating χ^2 , supported by G-test and Fisher's Exact test (Chapter Four, Section 4.2.1 and Legg, 2004), for statistical significance. This method of analysis would also show if there were any significant differences between the germination patterns of the nine species and all other species germinating from the seedbank.

Canonical Correspondence Analysis (CCA), using the CANOCO 4.5 program (ter Braak and Šmilauer, 2002) determined whether the variation in morphological measurements of the nine species could be explained by the physical and chemical environmental data recorded from the sampled 50 quadrat blocks (Chapter Four, Section 4.2). The species abundance data matrix that was part of the multivariate analysis described in Chapter Four (Section 4.2.3) was replaced with either a plant height matrix, number of flower-heads matrix or number of seeds produced matrix constructed from the morphological measurements collected in the same quadrat blocks (Section 5.3.1). A species presence absence matrix was added to the analyses as a covariable with the morphological data after an exploratory analysis (CCA) highlighted how strongly this factor was linked with environmental variables. A $\log_n$ transformation was performed on the seed matrix because of the extremely large inter- and intra specific variation in numbers of seeds recorded.

5.4 Results of field measurements

Individual representatives of the nine species were measured in the field to gauge variability in height (from substrate to highest part of plant). The numbers of each species were counted and recorded as were the numbers of flowering heads and seeds

produced per head (Section 5.3). Population density and seed production were calculated from the resulting data (Table 5.3).

5.4.1 Plant height

The numbers of plants that were measured for height varied between species because not all nine species grew in every quadrat block. Sixty individuals of *Tripleurospermum inodorum*, the least abundant of the nine species, were measured in 6 quadrat blocks and 210 individuals of *Holcus lanatus* were measured in 21 quadrat blocks. All of the plants measured were producing flowers and/or seeds and were therefore assumed not to be juveniles. There was considerable variation in the measured heights of individual plants within each of the nine species (Figure 5.6 and Table 5.3).

The two Fabaceae species, *Medicago lupulina* and *Trifolium campestre*, had similar median height, and were measured over a similar range of heights (Figure 5.6). The coefficient of variance (CV) of both species was high (approximately 80%) but the maximum measured height for *M. lupulina* in this study was only half of the published (usual) maximum (Table 5.3).

The two Asteraceae species, *Leucanthemum vulgare* and *Tripleurospermum inodorum* also had similar median height although *L. vulgare* was measured over a much greater range of heights (Figure 5.6). All of the *T. inodorum* plants measured were within the published range of usual measurements for the species and had the lowest CV of all nine species (39%) but *L. vulgare* was recorded at heights significantly different from expected, both higher and lower (Table 5.3).

Reseda luteola and *Senecio viscosus* were representative of the same bing habitat, although their morphologies are completely different. As expected *R. luteola* was by far the taller of the two species and demonstrated a greater coefficient of variance than *S. viscosus* (Table 5.3). The measured heights of *R. luteola* were significantly different from the published range, both higher and lower, but the measured heights of *S. viscosus* were significantly lower, with a maximum height of less than the mid point of the published range.

Table 5.3 Field results and calculations for the nine species

Height: The measured range of height for each species is compared with the published, usual range of height in the British Isles, taken from Stace (1997). The coefficient of variation (CV = 100 x standard deviation / mean) contrasts the degree of variance in measured height between the nine species

Density: Mean density for each of the nine species was calculated from the sum of the number of individuals counted in each quadrat block divided by the combined area of the 40 blocks (640 m^2)

Seed production: The minimum and maximum number of flower-heads per plant counted in the field and of seeds per flower-head is presented. The actual minimum and maximum number of seeds produced on any one plant was calculated for each of the nine species. The potential maximum seed production was calculated by multiplying the highest count of flower-heads recorded on any plant in the species by the highest count of seeds recorded on any flower-head in the species.

Species / Measurements	*Medicago lupulina*	*Trifolium campestre*	*Leucanthemum vulgare*	*Tripleurospermum inodorum*	*Reseda luteola*	*Senecio viscosus*	*Holcus lanatus*	*Centaurea nigra*	*Plantago lanceolata*
Range of measured height (mm)	30-400	10-350	20-980	140-430	110-1750	20-230	70-830	220-1220	50-890
Coefficient of variation (%)	79.5	80.4	83.1	38.9	87.6	68.7	69.4	50.4	74.7
Range of published height (mm)	50-800	20-300	200-750	100-600	500-1500	100-600	200-1000	150-1000	100-500
Density (m^{-2})	0.12	0.15	4.49	0.13	0.18	0.46	2.25	2.66	0.96
Flower-heads per plant									
minimum	18	1	1	1	1	1	1	1	1
maximum	723	64	51	14	19	37	71	411	21
Seeds per flower-head									
minimum	9	18	72	29	1187	29	33	27	3
maximum	33	44	340	93	1899	64	270	31	103
Seed production per plant (actual)									
minimum	162	18	72	29	1329	29	33	27	3
maximum	13737	500	8313	930	32718	882	4161	11508	1442
Potential maximum	*23859*	*2816*	*17340*	*1302*	*36081*	*2368*	*19170*	*12744*	*2163*

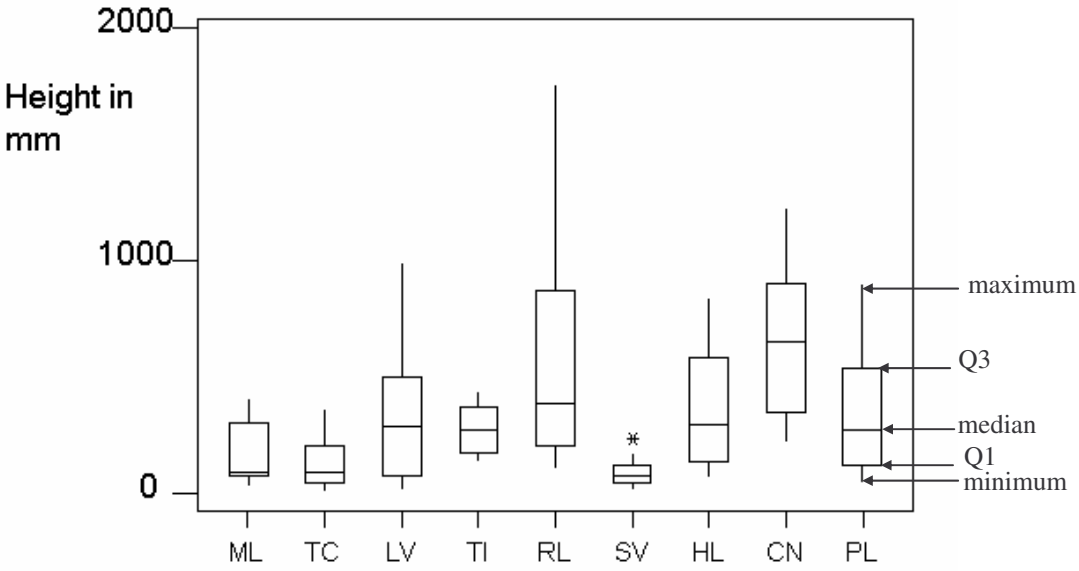

Figure 5.6 Height variation in the nine species

Boxplots showing the median, range and interquartile range (middle 50% of data, Q1-Q3) of height measured in each of the nine species. The asterisk (*) identifies an outlier in the data; a single measurement with a value lying between 1.5 and 3 times away from the middle 50% of the data (Minitab, Inc., 2000)
ML = Medicago lupulina, *TC* = Trifolium campestre, *LV* = Leucanthemum vulgare, *TI* = Tripleurospermum inodorum, *RL* = Reseda luteola, *SV* = Senecio viscosus, *HL* = Holcus lanatus, *CN* = Centaurea nigra, *PL* = Plantago lanceolata.

The three common species, *Holcus lanatus*, *Centaurea nigra* and *Plantago lanceolata*, displayed a similar inter-quartile range of height, although *C. nigra* measured taller than the other two species (Figure 5.6). The CV for *C. nigra* was 50%; considerably lower than the other species, except *T. inodorum*. When compared with the published range of usual heights, *H. lanatus* measured lower than or within the lower ranges, *C. nigra* measured higher or within the higher ranges, and although the interquartile range of *P. lanceolata* measurements were within the published range, there were both higher and lower measured heights (Table 5.3).

5.4.2 Density

There was considerable variation in the population densities of each of the nine species (Table 5.3) from one individual every 7 - 8 square metres, *Trifolium campestre, Medicago lupulina* (both Fabaceae) and *Tripleurospermum inodorum*

(Asteraceae), to one individual every 0.22 square metres, *Leucanthemum vulgare* (Asteraceae). *Senecio viscosus*, one individual every 2.17 square metres, was more than twice as densely distributed as *Reseda luteola*, one individual every 5.5 square metres. Individuals from the group of common species, *Holcus lanatus, Centaurea nigra* and *Plantago lanceolata*, were recorded at one individual every 1- 0.4 square metres, more frequently distributed than four of the other species but less frequent than *L. vulgare*.

5.4.3 Seed production

Flower-head and seed production in each of the nine species varied considerably both between and within species (Table 5.3). The minimum counted seed production per plant in all nine species is between 0.2 - 5.6 % of the maximum counted seed production per plant. The potential maximum seed production was calculated by multiplying the highest count of flower-heads on any plant by the highest count of seeds on any flower-head and was always greater than the actual seed production. The difference between potential maximum and counted (actual) maximum numbers of seed produced by a single flower-head varied by less than 5% in *Centaurea nigra,* but was more than 80% in *Trifolium campestre*.

5.5 Results of trials

A series of glasshouse trials were set up as described in Section 5.3.3 to establish the viability of seeds within each of the nine species, measured as percentage germination success, and to ascertain the availability of all species in the seedbank, measured as the number of emerging seedlings from substrate samples.

5.5.1 Glasshouse germination trial

Germination rate was consistently higher in the five perennial species than the four annual species. More than 30% of the 240 sown seeds of *Leucanthemum vulgare*, *Reseda luteola*, *Holcus lanatus*, *Centaurea nigra* and *Plantago lanceolata* germinated successfully (Figure 5.7).

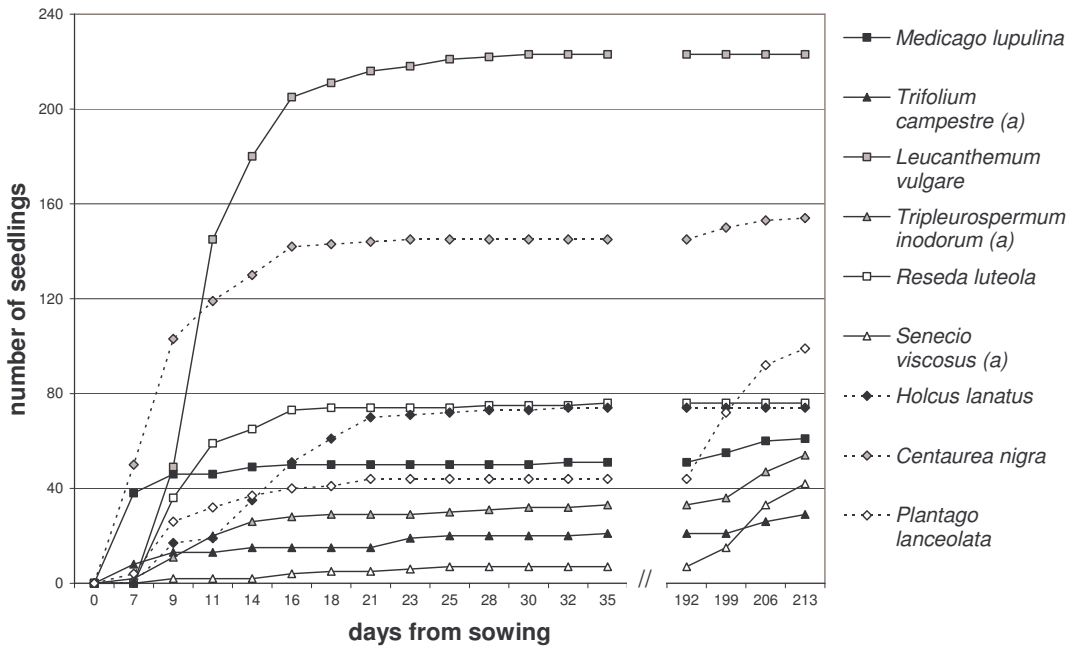

Figure 5.7 Germination trial

Two hundred and forty seeds of each species were sown in the mist room of the glasshouse on 11/10/99, day 0. The first seedling count was 18/10/99, day 7, then every Monday, Wednesday and Friday for seven weeks. The seed trays were moved to the cold frame and monitored weekly. // on the x-axis denotes a break in the displayed data when there was no germination recorded in any of the species (between 17/11/99 and 22/04/00). Germination continued on day 199 after a series of hard frosts toward the end of April 2000. There was no further germination after 13/05/00, day 213, although weekly monitoring continued until August. (a) after the species name highlights the annual species.

The germination rate in *L. vulgare* was very high, 93%, and all of the seeds that germinated did so during the initial 10-week period in the glasshouse. Successful germination of *R. luteola* (32%) and *H. lanatus* (31%) also took place completely within the early stages of the trial. Initial seedling emergence in all three of these species was slightly slower than for most of the others and the first seedlings were recorded on the ninth day after sowing although germination was very rapid over the following week. There was no further germination in the cold frame.

Germination in the remaining six species occurred both in the glasshouse and, after severe frost, in the cold frame. The two perennials displayed very different patterns of emergence. One hundred and three of the *C. nigra* seedlings emerged within nine

days and a further 42 in the first three weeks in the glass house. The remaining nine seedlings emerged in the cold frame resulting in a total germination rate of 64%. In contrast only 44 of the *P. lanceolata* seeds germinated in the glass house. The remaining 55 seedlings emerged rapidly after the frost resulting in a total germination rate of 41%.

As already mentioned germination success in the annuals was much lower. Thirty-eight of the germinable *Medicago lupulina* seeds germinated within the first seven days then germination was very slow for the remainder of the glass house stage of the trial. The frost-induced burst of activity six months later, in the following April resulted in a germination success of only 25%. *Trifolium campestre*, the other Fabaceae, was the least successful of the nine species with only 12% of seeds germinating. Many of these seedlings emerged within the first seven days but germination occurred in apparently random small bursts throughout the glasshouse stage of the experiment and the cold frame germination began a week later than the other species. *Tripleurospermum inodorum* also began to emerge within the first seven days and the rate of germination increased over the next two recording periods, then dropped to only occasional single seedlings. Further germination over the three weeks resulted in 23% germination success. *Senecio viscosus* had a very slow emergence rate in the glass house, only seven of the 240 sown seeds. A further 35 seeds germinated in the cold frame (18% germination success).

5.5.2 Soil seedbank trial

During the three-month observation period 514 seedlings from 46 species were identified and recorded. No ungerminated seeds were found in the samples at the end of the trial and there were no seedlings recorded in the control trays. Exactly 200 seedlings were from the nine species in this study (Table 5.4).

Holcus lanatus and *Plantago lanceolata* germinated from surface and unweathered samples at all three recording sites although *Centaurea nigra* was not recorded in any samples from any sites. *Leucanthemum vulgare* and *Reseda luteola* germinated in almost equal numbers from both the surface samples and from the unweathered substrate but the other species were more abundant in the surface samples. It should

be noted that all but one of the *L. vulgare* and *R. luteola* seedlings emerged from samples collected from Greendykes.

Table 5.4 Soil seedbank trial

The numbers of seedlings of each of the nine selected species germinating from 40 surface soil samples and 40 unweathered substrate samples (10 cm diameter and 5 cm deep) collected from each of the three bing sites are presented with the total number of seedlings of each species recorded and number of surface and unweathered samples containing the seedlings. The same data is presented for seedlings of other species and for all species emerging, including the nine, for comparison.

OB = Oakbank; CL = Clapperton; GD = Greendykes; sur=surface soil samples; unw = unweathered substrate samples

species	number of seedlings from surface soil			number of seedlings from unweathered substrate			total number of seedlings	number of samples with germinating seedlings	
	OB	CL	GD	OB	CL	GD		sur	unw
Medicago lupulina	1	0	0	1	0	0	2	1	1
Trifolium campestre	18	0	0	3	0	0	21	4	2
Leucanthemum vulgare	0	0	27	0	1	28	56	8	11
Tripleurospermum inodorum	0	3	0	0	0	0	3	3	0
Reseda luteola	0	0	12	0	0	11	23	4	3
Senecio viscosus	0	0	0	0	1	0	1	0	1
Holcus lanatus	15	10	3	5	5	2	40	8	9
Centaurea nigra	0	0	0	0	0	0	0	0	0
Plantago lanceolata	45	2	1	3	0	3	54	9	5
Other species	119	44	54	32	20	45	314	33	27
ALL SPECIES	198	59	97	44	27	89	514	34	31

Of the annual species, *Trifolium campestre* had the largest number of seedlings germinating from the seedbank with 18 seedlings sprouting from the surface samples and three from the unweathered substrate collected at Oakbank bing. Three seedlings of *Tripleurospermum inodorum* germinated from the surface samples collected at Clapperton bing and only two seedlings of *Medicago lupulina* germinated, one from the surface samples and one from the unweathered substrate collected at Oakbank bing.

The inter-specific variation in numbers and distribution of seedlings between the surface and unweathered samples was significant but there was no significant difference between the numbers of samples with germinating seedlings (calculating

χ^2 as described in section 5.3.6). There was also no significant difference between the combined total seedlings from the nine species and the seedlings from all other species between the surface and unweathered samples.

The overall seedling emergence was different in each of the three sampled bing sites (All species, Table 5.4). There were similar numbers of germinating seedlings in the surface and unweathered samples taken from Greendykes bing but recorded emergence was much higher from the surface samples than the unweathered substrate at Oakbank and Clapperton. The total number of species germinating from samples taken from Clapperton was significantly lower than from the other two sites (χ^2). However more complex statistical analyses of seedbank data such as these is not considered to be appropriate due to the very small counts of some species and the many zero values in the data (Bakker *et al*., 1996).

5.6 Analyses of combined measurements and data collection

The data resulting from the field measurements and glasshouse trials were analysed in accordance with the methods described in Section 5.3.6. Correlations of the field and trial measurements were calculated between and within each of the nine species. The data were also analysed by Canonical Correspondence Analysis (CCA) in conjunction with the physical and chemical environmental data that was recorded from the fixed quadrats and analysed with all species distribution and abundance in Chapter Four (Section 4.5).

5.6.1 Correlations and calculations

Correlations between the field measurements of each species were expected to comply with the model: the taller the plant the more flower-heads; the more flower-heads the more seeds on each flower-head; the more seeds the greater the number of plants (population density); the greater the abundance the taller the plants (due to competition for light).

The correlations between measured data from *Tripleurospermum inodorum* plants exemplified this popular view of vigorous growth and copious seed production in pioneering or colonising species and show strong positive relationships between

these physiological measurements (Table 5.5). However none of the other species displayed all of these associations and a mixture of positive and negative correlations of varying scales was apparent for each of the paired data sets.

Table 5.5 Relationships between physiological measurements

The correlation coefficient was calculated for every combination of measured data collected in the 40 quadrat blocks from each of the nine species. The correlations highlighted in bold emphasise strong positive relationships.

ht = mean heights of individual plants; ab = numbers of individuals per quadrat block (population density); flo = mean numbers of flowering heads per plant; sd = mean numbers of seeds per flowering head.

species	ht:ab	ht:flo	ht:sd	ab:flo	ab:sd	flo:sd
Medicago lupulina	0.605	0.564	0.227	-0.612	-0.522	0.313
Trifolium campestre	-0.253	**0.771**	0.330	0.139	-0.052	-0.159
Leucanthemum vulgare	-0.555	0.671	-0.215	-0.240	**0.932**	-0.051
Tripleurospermum inodorum	**0.739**	**0.855**	**0.982**	**0.850**	**0.987**	**0.983**
Reseda luteola	-0.432	0.377	0.068	0.578	0.054	0.014
Senecio viscosus	**-0.949**	**0.958**	0.171	**-0.976**	-0.098	0.047
Holcus lanatus	0.468	0.480	-0.039	0.352	-0.357	**0.790**
Centaurea nigra	-0.664	**0.788**	-0.097	-0.092	-0.052	-0.122
Plantago lanceolata	-0.217	0.647	0.536	0.138	-0.262	0.079

The individual plant height to number of flower-heads ratio (ht:flo) showed a positive correlation in all nine species but this is likely to be a function of the modular growth plan of all vascular plants. None of the other ratios were consistently positive or negative for all nine species. For example *T. inodorum* plants were taller with increasing abundance but *S. viscosus* plants were smaller. There was little relationship between the number of flower-heads produced on each plant and the number of seeds produced on each flower-head on *Leucanthemum vulgare* and *S. viscosus* plants, yet on both *T. inodorum* and *Holcus lanatus* plants there was a strong positive correlation between these two sets of data.

The variation in measurements relating to seed ecology between species and between stages of development is summarised in Table 5.6. The loss of individuals at each stage of development is calculated by combining the results of field measurements and germination trials. There are clear discrepancies between the expected density of plants calculated from the germinable seedbank and the population densities recorded in the field.

Table 5.6 Summary of physiological results

Total seed production per m^2 (the sum of flowering heads x seeds per flower-head recorded in the field on each individual plant /640), germinable seed production per m^2 (percentage germination success in the glasshouse trial x total seed production), germinable seedbank per m^2 (extrapolated from the number of seedlings emerging in the seedbank trial) and density of plants per m^2 (from table 5.3.) have been presented for each of the nine species.

species	total seed production	germinable seed production	germinable seedbank	density of plants (measured)
Medicago lupulina	430	108	2	0.12
Trifolium campestre	108	13	21	0.15
Leucanthemum vulgare	2,546	2,368	56	4.49
Tripleurospermum inodorum	116	27	3	0.13
Reseda luteola	3,154	1,009	23	0.18
Senecio viscosus	224	40	1	0.46
Holcus lanatus	6,995	2,168	40	2.25
Centaurea nigra	1,835	1,174	0	2.66
Plantago lanceolata	297	122	54	0.96

5.6.2 Relationships to physical and chemical environment

Canonical Correspondence Analysis (CCA) of the individual components of the measured physiological data for the nine species (height, density, number of flower-heads and seed production) with physical environmental data was calculated with supplementary chemical environmental data. The influence of each of the environmental factors on the abundance of the nine species has already been demonstrated within the data for the most abundant species (Chapter Four, Figures 4.5 and 4.8).

The amount of explained variance in the CCAs of the physiological data with the same environmental factors ranged from 14% of the 14% variation in seed production, attributed to the chemical environment (< 2% explained variance), to 58% of the 66% variation in flower production, also attributed to the chemical environment (≈ 38% explained variance).

An exploratory CCA demonstrated that the percentage of explained variance was strongly influenced by the presence and absence of species within the environmental gradients. Variation of presence/absence in the species data was described by both

the physical (45.84%) and the chemical (32.84%) environmental variables and 45.3% of this variance was described by the first four constrained axes. The graphical display of the analysis as a triplot (Figure 5.8) presents the main influences of the physical environment on species presence or absence as bare ground and management and of the chemical environment as pH, sodium and potassium. The distribution of the nine species names on the triplot represents the extent that the presence or absence of each species is influenced by the environmental variables: for example *Tripleurospermum inodorum and Medicago lupulina* are positioned between the management and pH axes indicating that their presence is linked with both increased pH and management.

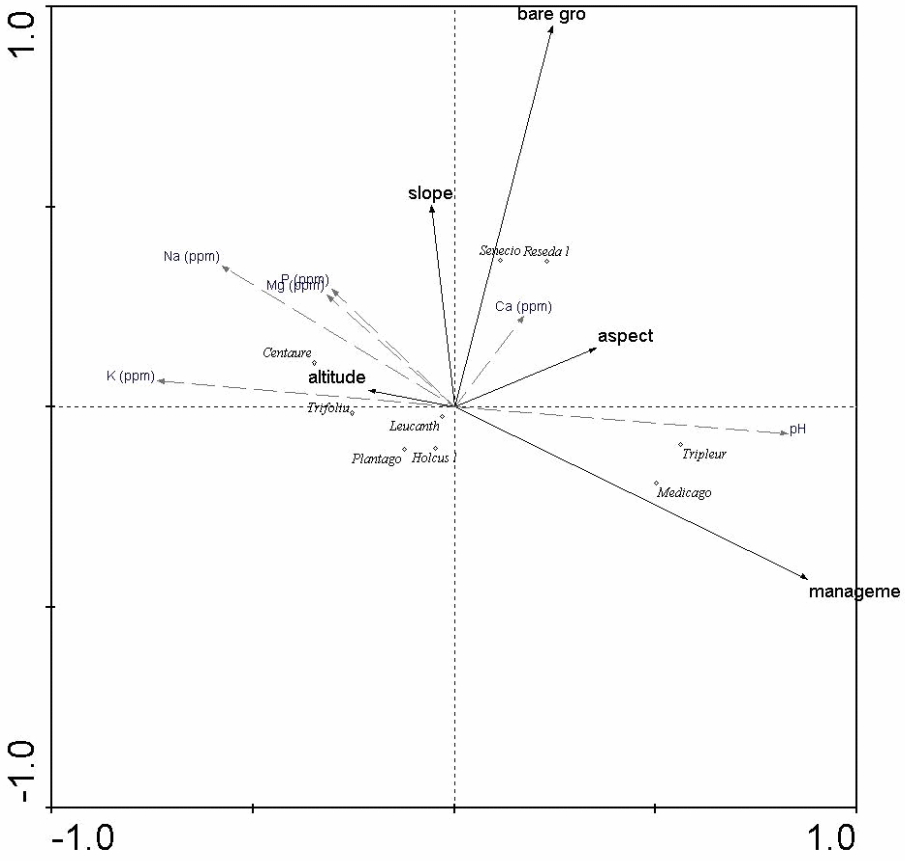

Figure 5.8 Triplot representing the CCA of species presence/absence with environmental variables

Because of the high percentage of variance in the species data that was explained by presence/absence the decision was made to repeat the analyses including presence/absence as a covariable within each CCA. The percentage of variance in the measured plant attributes that is described by the presence/absence covariable is unique to each of the analyses (Table 5.7). The inclusion of the covariable in the analyses reduces the percentage variance in the species data that is described by the environmental data (Phys% and Chem% in Table 5.7) by removing the effect of zero measurements when there were no representatives of a species present in the recording block. This change in emphasis can be demonstrated by comparing the triplot graphs of the CCA of the species flower-head matrix with environmental variables only (Figure 5.9) and with the presence/absence covariable included in the analysis (Figure 5.10).

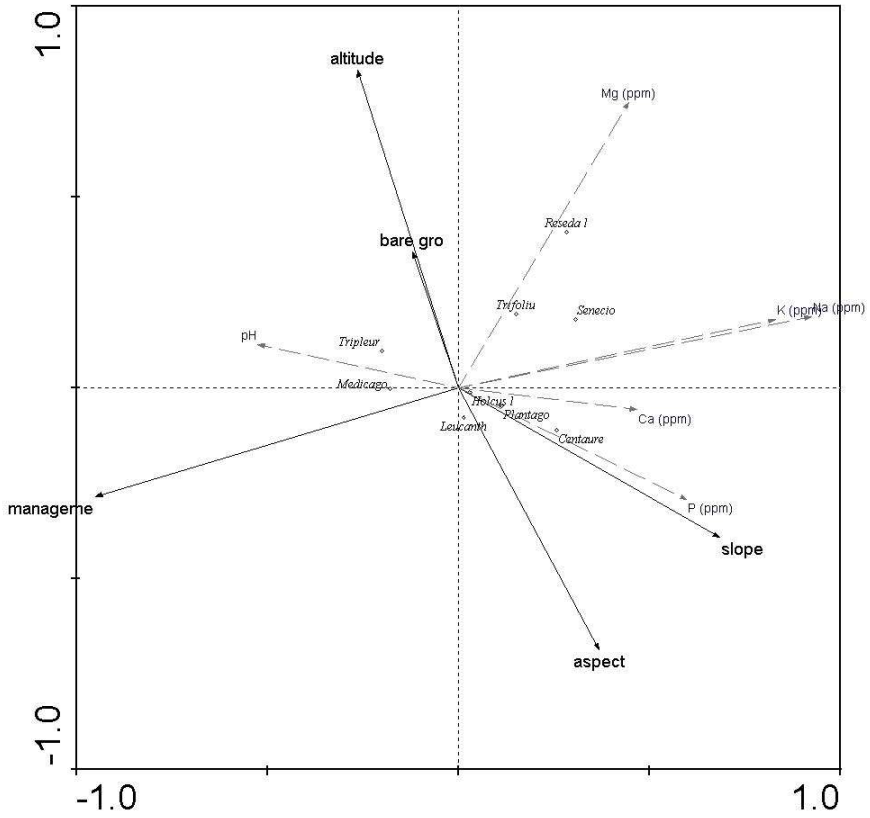

Figure 5.9 Triplot representing the CCA of species flower-head production with environmental variables

The relationship between this figure and Figure 5.10 are discussed in the text

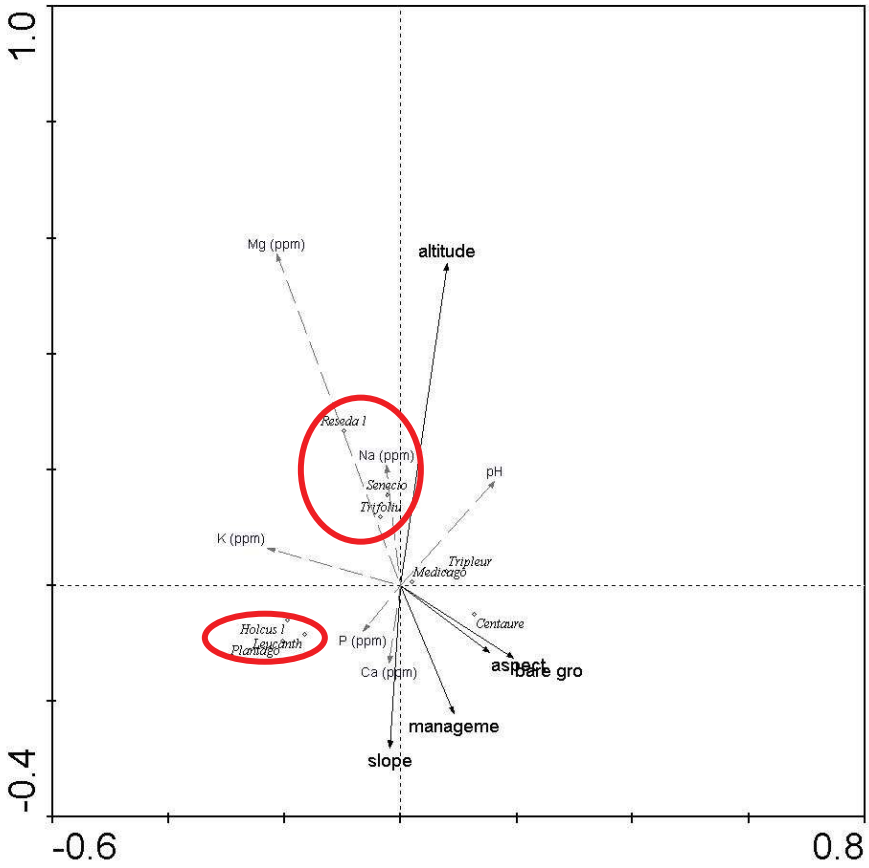

Figure 5.10 Triplot representing the CCA of species flower-head production with environmental variables and presence/absence covariable

The relationship between the environmental variables and the two groups of species circled in bold are discussed in the text. The reduction in axis length of management, aspect and slope between this figure and Figure 5.9 demonstrated that the variance on the number of flower-heads produced, that was apparently explained by these factors, was mainly due to the presence or absence of species.

The analysis without presence/absence covariable (Figure 5.9) was difficult to explain and variation in the number of flower-heads produced by *Holcus lanatus*, *Plantago lanceolata* and *Leucanthemum vulgare* plants could be interpreted as correlated positively with phosphorus, aspect and/or slope, or negatively correlated with altitude and/or pH. When the covariable was included in the analysis (Figure 5.10) these three species were grouped more closely together and the production of flower-heads was shown to be more clearly negatively correlated with the pH axis. Altitude and increased magnesium in the substrate were shown to correlate with increased flower-head production in *Reseda luteola*, *Senecio viscosus* and *Trifolium*

campestre both with and without the presence/absence covariable being included in the analysis. The log report of the CCA, produced by the CANOCO programme, confirmed that altitude (0.9249 correlation) and magnesium (0.6853 correlation) corresponded with the second constrained axis. The result of including the covariable in the analysis of flower-head production was to describe more than half of the variance in relation to presence of absence of species recorded in the quadrat blocks and in doing so reducing the amount of variance described by the physical and chemical environmental variables. The percentage of the variance that was now described in the first four constrained axes increased from 58.4% to 71.3%.

Increased heights in individual plants of the nine species were each correlated with different environmental factors. Plasticity (measured as variation in height) was linked to both the physical environment and the availability of nutrients. Interpretation of the CCA, without covariables, suggested that the height of *Medicago lupulina* plants was positively correlated with increased management and heights of *Holcus lanatus*, *Plantago lanceolata* and *Leucanthemum vulgare* plants were negatively correlated with altitude. However when the presence/absence covariable was included in the analysis the heights of *M. lupulina* and *H. lanatus* plants were now positively correlated, and of *Tripleurospermum inodorum* plants strongly negatively correlated, with calcium. More than 50% of the variance in height explained in the first analysis was apparently describing the presence and absence of species in the recording quadrats.

Variance in seed production was the least well explained by the measured environmental variables. A CCA of seed production with environmental variables including the presence/absence covariable suggested that variance in seed production in the nine species was not strongly correlated with any of the measured environmental factors and a comparatively low percentage of the variability was described. On reviewing the raw data the range of variation in numbers of seed produced both between and within species was noted (the highest counts were often more than 1,000 times greater than the lowest) and the decision was made to reanalyse the variables using the natural logarithm ($\log_n$) of seed numbers.

Transforming the data did not clarify the associations between seed production and the environmental variables.

In all of the analyses, both with and without the presence absence covariable, the first two constrained axes described most of the variance (Table 5.7). The variance in any of the species data that is explained by the environmental variables is the percentage variance calculated from the total inertia and eigenvalues given in the summary of results produced in the log (printed record) of the analysis (described in Appendix 16). The cumulative percentage variance is the amount of the calculated variance that is described by the first four constrained axes in the analyses.

Table 5.7 Summary of CCA results

The cumulative percentage variance gives the amount of variance explained by the first four constrained axes of the current environmental variables. The total inertia (the sum of the eigenvalues for the same four axes including any covariables), and the sum of all eigenvalues (EV) for only the environmental variables are used to calculate the percentage variance in the species data that is explained by the covariable (Cov %), the physical environmental variables (Phys %) and the chemical environmental variables (Chem %). A more complete explanation of the calculations and an example of analysis results are given in Appendix 16.

Species data	Cumulative percentage variance 1st axis	2nd axis	3rd axis	4th axis	Total inertia	Sum of all EV	Cov %	Phys %	Chem %
Presence absence	29.3	42.5	44.2	45.3	1.071	1.071	N/A	45.84	32.84
Height	22.8	38.0	40.3	41.1	0.953	0.953	N/A	41.44	43.55
Height + covariable	21.6	36.1	39.4	40.1	0.953	0.444	53.41	18.88	28.86
Flowers	41.1	52.8	56.6	58.4	1.330	1.330	N/A	58.96	65.86
Flowers + covariable	44.4	65.5	69.1	71.3	1.330	0.519	53.46	28.05	29.92
Seeds	13.2	14.0	14.1	14.1	2.769	2.769	N/A	14.08	18.02
Seeds + covariable	3.4	5.0	5.8	6.1	2.769	2.032	26.62	4.51	9.61
Log_n seed + covariable	8.4	11.5	13.2	14.4	2.278	1.431	37.18	9.35	17.47

5.7 Discussion and conclusions

The results from the field measurements and glasshouse trials demonstrated the inter- and intra-specific variation of the nine species and established that individual morphological features associated differently in each species. There is no simple model that can be established to describe the processes of reproduction, distribution and establishment of colonising species by extrapolating the information from these data.

5.7.1 Morphological measurements

The height ranges of individual plants in three of the species measured both significantly lower and higher than the published (Stace, 1997) usual ranges of height: *Leucanthemum vulgare*, *Reseda luteola* and *Plantago lanceolata*. Smaller plants are perhaps understandable, due to the inhospitable conditions on the surface of the bings, where there is no protection from the elements. Taller plants usually occur when there is competition for light or a nutrient rich soil. This has been demonstrated in tree species by King, (1991) and Koch *et al*. (2004). Variability in height in species growing on the bings may be an element of plasticity that is retained in early successional species to ensure that tall individuals, capable of more effective distribution, are available when environmental conditions are suitable. When flower-heads are positioned high on the plant, as they are in these species, the distance of dispersal of seeds from the adult plant is greater in a tall plant than a short plant. Carefully designed (and extensive) field trials would be required to establish the validity of this theory.

5.7.2 Germination trials

The germination trials produced results that were dissimilar to those reported in the much larger trials conducted by Grime *et al*. (1981). Although different experimental techniques were used to determine germinability in that study, the specimens were collected from similar substrates; limestone outcrops and disused quarries.

The total percentage germination and time to 50% germination (t_{50}) were compared with the findings of Grime *et al.* (1981) for 'germination of freshly collected seeds' of the nine species (Table 5.7). There were significant differences in either the total germination success or the number of days taken to achieve 50% of the successful germination (or both) in all species apart from *Leucanthemum vulgare.* The total percentages of successful germination in *Leucanthemum vulgare* and *Plantago lanceolata* were comparable with those recorded by Grime. The percentage of *Medicago lupulina* and *Centaurea nigra* seedling eruption was significantly higher in the trials conducted in this study. Conversely, successful germination of *Trifolium campestre*, *Tripleurospermum inodorum*, *Holcus lanatus* and *Reseda luteola* was significantly lower. *Senecio viscosus* germination appears to be much lower in the Sheffield study but the 1% success reported is for immediate sowing (without chilling) and is probably more comparable with the glass house section of my trial.

Table 5.8 Comparison of germination results

The germination results for the nine species recorded in this study are compared with those from the study of the Sheffield flora (Grime et al.*, 1981). The number of seedlings germinating from 240 seeds (Figure 5.7) is converted to percentage germination and t_{50} is the time in days required for 50% of the viable seeds to germinate.*

species	*Medicago lupulina*	*Trifolium campestre*	*Leucanthemum vulgare*	*Tripleurospermum inodorum*	*Reseda luteola*	*Senecio viscosus*	*Holcus lanatus*	*Centaurea nigra*	*Plantago lanceolata*
total germination (%)	25	12	93	23	32	18	31	64	41
published germination (%)	5	72	92	72	80	1	100	36	37
t_{50} (days)	<7	15	12	17	12	>200	17	10	>200
published t_{50} (days)	7	5	8	21	4	>200	3	35	14

The times to 50% germination (t_{50}) for seeds of *T. inodorum, L. vulgare*, *S. viscosus* and *M. lupulina* were comparable with the Sheffield study. Germination of

T. campestre, *R. luteola*, *H. lanatus* and *P. lanceolata* seeds was significantly slower than reported and *C. nigra* seedlings emerged more rapidly. *P. lanceolata* emergence from the bing substrate was more successful after the April frost.

The double germination pattern of six of the nine species, the species in the trials from this study, could have been examined as two separate germination events and would have made the results more comparable with the Sheffield study but would not be so representative of field conditions, which are ultimately what is being simulated. The combined trial with a single cohort of seeds demonstrates that within a single growing season there are species, like *Plantago lanceolata*, that produce some seeds with the inherent plasticity to germinate immediately while the non-germinating seeds remain dormant until particular stimuli are received (in this case severe chilling). Other species produce seeds that are all 'programmed' for immediate germination. In this study these were not annual species and the most extreme example was *Leucanthemum vulgare* with 93% germination from freshly sown seeds (a finding comparable with Grime *et al*., 1981). Yet other species produce seeds that remain dormant until activated by a stimulus other than cold. The sporadic germination of *Trifolium campestre* seeds possibly exemplifies this last mechanism.

The lack of similarity between individual species in the two sets of trials (this study and Grime *et al*., 1981) could be linked to differences in the provenances of the seeds being used. Mkonda *et al*. (2003) demonstrated that cumulative germination percentage differed significantly between provenances in seeds of *Strychnos cocculoides,* a wild indigenous fruit tree of Zambia.

In addition climatic conditions in central Scotland and the Sheffield area are quite different and this alone could lead to plants producing seeds that respond to different germination stimuli. Mean climate data over the 30 years from 1971 – 2000 show that, in comparison with Edinburgh, Sheffield has 20 fewer days of frost annually, higher minimum and maximum temperatures throughout the year, and higher rainfall and more rain days every month of the year except July (Meteorological Office, 2004a; 2004b).

The simple germination trial of this study could be replicated using seed collected from cohorts of the same species growing in the same sites in different years or using seeds collected from cohorts of the same species growing in different conditions (Cornwall and Caithness for example) in the same years. The resulting information would give a clearer understanding of the extent of plasticity and variability in this one small contributing factor to species distribution. Many of the experimental conditions produced in the Sheffield trials are not representative of normal field conditions in either Sheffield or central Scotland (e.g. dry storage for 3 and 6 months, or moist storage at 20^{o}C). There is, however, potential for more, detailed trials on the effects of moist storage at lower temperatures on the germination of seeds.

5.7.3 Seedbank trials

The total number of seedlings, from all species, emerging from the seedbank on the bings was very low compared to numbers recorded in abandoned wet meadows by Jensen (1998); however the literature suggests that these results are an accurate representation of the seedbank. The bulking of samples increased the likelihood of species being represented in the sample. Reduction of substrate bulk is recommended in seedbank analysis as the number of species emerging is higher in concentrated samples and the process of reduction scarifies the seed coats (ter Heerdt *et al*., 1996; Thompson *et al*., 1997). Traba *et al*. (1998) suggest that there may be a loss of species with seed diameters less than 0.2 mm in concentrated samples and although this may prevent a few of the total number of species germinating from the seedbank it should not affect the volume of emergence from the nine species in this study as their seeds all have a larger diameter (even *R. luteola*).

Glasshouse experiments have been shown to result in higher germination rates than field trials for arable weeds (Roberts & Ricketts, 1970; Graham & Hutchings, 1988) and it would have been advantageous to replicate the germination trials in the field. Unfortunately the agreements to carry out research on the bings precluded manipulative experiments or *in situ* trials. Only Seafield, which has also been utilised as a landfill site (Appendix 1), could have been used for this purpose but was not comparable with any of the other bing sites.

More than half of the seedlings that emerged from the Greendykes samples grew from the unweathered substrate, 5 cm below the surface. This appears to confirm that some species, particularly *Leucanthemum vulgare* and *Reseda luteola*, can persist through enforced dormancy when seeds become buried. A key feature of seedbanks is the ability of seeds from some species to survive in a dormant state for 20 years or more until conditions for germination are suitable; indeed Ramsbottom (1942) describes the germination of an herbarium seed that was known to be 237 years old. Thompson *et al*. (1997) make a general inference that seeds buried at depth are older than seeds on the surface and represent non-transient seed banks, a suggestion that seems to be supported by the evidence collected from the nine selected species in this study. Despite four of the nine species being annuals only *Tripleurospermum inodorum* was recorded solely from surface samples suggesting that a persistent seedbank may be retained in the bing substrate. This theory could be investigated further using the data collected for other species germinating from the samples and comparing them with the above ground vegetation data, however the coarse structure of the bing substrate must also be taken into account as it will allow all but the very largest seeds to be washed between the shale particles, probably to considerable depths.

5.7.4 Multivariate analyses

Morphological variation and seed production between and within species was explained in relation to the physical and chemical environment in a series of Canonical Correspondence analyses. Once the confounding influence of presence/absence was removed from the analyses it became clear that the variance in height, flower production and seed production of each of the nine species was explained by different environmental variables.

There was a positive correlation between the height of *Medicago lupulina* and calcium yet this species was consistently measured at less than 50% the published height (Stace, 1997). There were no quadrats where available calcium was measured as deficient in the substrate (Bradshaw and Chadwick, 1980) that might explain lack of growth. The species is acknowledged to be recorded most frequently in alkaline

soils (Hill *et al*., 1999) and conditions on the bing substrate are alkaline. *Tripleurospermum inodorum* plants were also consistently smaller than the published expected height but the height of this species is negatively correlated with calcium. It would be imprudent to emphasise the importance of this information however as less than 11% of the total variance in height of all nine species is explained by the combined influence of the six measured variables of the chemical environment.

The comparatively high percentage of explained variance in flower production was unexpected and some of the correlations with environmental variables were very strong. Increased flower production in *Reseda luteola, Senecio viscosus* and *Trifolium campestre* plants was positively correlated with magnesium, sodium and altitude but negatively correlated with management. In *Holcus lanatus*, *Leucanthemum vulgare* and *Plantago lanceolata* plants flower production was negatively correlated with pH and positively with potassium and phosphorus, but the opposite was true of *Tripleurospermum inodorum* and *Medicago lupulina* plants. Increased flower production in *Centaurea nigra* plants correlated positively with increased bare ground and negatively with potassium. Fabbro and Korner (2004) documented a decrease in the number of flowering heads produced by individual plants, with increased altitude in alpine habitats: a relationship that was not evident in this study, possibly due to the comparatively limited variation in altitude on shale bings.

The seemingly strong influence of the environment on flower production in all nine species made the lack of explained variance in seed production by the measured environmental variables difficult to interpret. In some instances a variable that had a positive correlation with flower production had a negative correlation with seed production in the same species or *vice versa*: for example magnesium, positive for flower production and negative for seed production in *Trifolium campestre,* and phosphorus, negative for flower production and positive for seed production in *Tripleurospermum inodorum.* The lack of strong positive correlation between number of flowering heads and numbers of seeds produced was demonstrated earlier in this chapter but does not explain the anomaly of a single environmental factor having apparently opposing effects on the fecundity of a species.

I can find no references from literature that either support the findings from these analyses or offer alternative theories for the causes of the extreme vagaries of within and between species physiology in these nine species.

5.7.5 The nine species

The two Fabaceae were selected because despite their similarity of physiology and habitat requirements, they were not recorded together on the bings. The two species were measured at similar overall population densities on the bing sites but the relationship between the other measurements recorded from the two species was very different. The height of *M. lupulina* plants increased with abundance while the number of flower-heads and number of seeds per flower-head both decreased but there was little correlation between the same measurements in *T. campestre* plants.

The presence of *Medicago lupulina* in the seedbank of both the surface soil and unweathered substrate at Oakbank was not anticipated, as it was not recorded as a surviving plant in any of the quadrats from this site. There must be, or have been, a local seed source, perhaps the surrounding agricultural land. Pavone & Reader (1982) found that seedbank size in *M. lupulina* ranges from a few tens per m^2 to several thousand, depending on topography, habitat and past history. Reader (1993) recorded that predation in or on the ground accounted for the loss of 80% of *M. lupulina* seeds in abandoned pasture. While this may go some way towards explaining the lack of recorded plants at Oakbank it does not fully explain the absence of germination from the substrate samples of sites where *M. lupulina* plants were producing plentiful, viable seed. Some factor(s), not considered in this investigation, is preventing the germination or establishment of this species.

Trifolium campestre was found in high numbers in the seedbank, compared to the other annual species, and only at Oakbank. It was recorded in both the surface soil and the unweathered substrate so can be considered to be persistent on this site. The germination density in the seedbank trial was higher than that expected from the results of the germination trial. It is possible that the apparently random bursts of germination noted in the trial continue throughout the year as a response to different

stimuli and that the potential germination for this species was under recorded as a result.

Leucanthemum vulgare and *Tripleurospermum inodorum* were also selected as a pair of species, similar in appearance and from the same family. The glasshouse trials established that over 90% of *Leucanthemum vulgare* seeds, collected from seedheads, germinated immediately with no further germination after chilling, indicating an absence of any innate dormancy. Grime *et al*. (1981) reported similar results (90-100%) in their trials. The recorded density of this species in the field is high, but this is not unexpected as *L. vulgare* is a perennial species that persists above the ground rather than below and is also more competitive than many other species. These strategies allow sustained survival in undisturbed sites.

Fenner *et al*. (2002) demonstrated that, for both *L. vulgare* and *T. inodorum*, predation on the seeds in the capitulum increases with size of capitulum and that *L. vulgare* consistently has larger capitula (an observation supported by the measurements in this study). Studies of the two species in populations from grassland, arable fields and waste ground in Hampshire (Fenner *et al*., 2002) revealed that percentage insect infestation suffered by *T. inodorum*, 33% (±12.3), is higher than the 26% (±11.8) recorded in *L. vulgare*. The authors have assumed that seed infestation is synonymous with predation. Post-dispersal seed predation is also very high for many species. In a study of abandoned pasture Reader (1993) suggests that up to 93% of *L. vulgare* seeds may be lost to predation when in or on the ground.

A combination of the two types of predation would considerably reduce the number of seeds being retained in the seedbank and may explain the apparent lack of seeds in the seedbank trials. The 93% losses to predation presented by Reader (1993) would reduce the *L. vulgare* potential seedbank from 2,546 per m^2 (mean seeds produced) to 16 per m^2 (seeds remaining after pre- and post-dispersal predation), considerably closer to the actual density of plants recorded in the field than germination and seedbank trials would infer. *Tripleurospermum inodorum* seedlings emerged only from surface soil samples. Although the numbers of individuals recorded are small, this suggests that either the species has colonised the area recently, such that the

seeds have not yet been moved through the spaces between particles in the substrate, or that it is a transient species as described by Thompson *et al*. (1997). The species is certainly recognised as a ruderal that is often found in cultivated ground (Smith *et al*., 2002) and a requirement for high levels of disturbance may limit its distribution on the bings and explain why it is not recorded consistently with any of the other species.

Reseda luteola and *Senecio viscosus* were two unrelated species apparently sharing the same ecological niche and abiotic resources. Both species are strongly linked with a high percentage of bare ground suggesting that they are not in competition for any resources. They also represent the mixture of longer-lived and shorter-lived pioneers that are common to shingle banks (Scott, 1963; Grubb, 1987). On the bings *Reseda luteola* and *Senecio viscosus* were representative of the vegetation on a scree-like habitat that is not found in the surrounding landscape. *S. viscosus* is a 'conventional' short-lived, annual, pioneer species whose appearance on the bings reflects a strong association with man-made sites and transport routes. There is considerable debate as to whether it is native. Smith *et al*. (2002) record the species as native, Stace (1997) states that it may be native and the New Atlas (Preston *et al*., 2002) suggests that the species is not native, except perhaps as a genetic dwarf form on coastal shingle. As previously noted, the specimens on the bings were consistently smaller than the expected normal range of height and the comparatively isolated examples on the bings may be genetically allied with the coastal shingle variants. Unfortunately Preston *et al*. (2002) do not define the native distribution. The very strong negative correlation between height and abundance, and positive correlation between height and number of flower-heads found in *S. viscosus* plants recorded on the bing sites may be indicative of a restricted gene pool caused by a combination of isolation and apomixis.

R. luteola is a longer-lived biennial whose native status is also debated. The seeds of this species are dispersed by wind, although they are released from capsules then roll along the ground rather than being dispersed through the air over long distances (Hodgson *et al*., 1995). Again it has strong associations with man-made sites but exactly how the seeds travel from one site to another is not clear. The measurements

recorded in this study showed no strong correlation in *R. luteola* although individuals demonstrated a larger than expected plasticity of height (Stace, 1997). *R. luteola* produced more than ten times as many seeds as *S. viscosus* and more of the seeds germinated from the seedbank in trials yet the population density of plants on the bing was much lower. Predation must play a considerable part on the survival of both seeds and seedlings, as it does in other species, and lack of suitable microsites for establishment may also be a contributing factor but I was unable to find any definite information on either of these species in the literature.

The three species selected because they are very common on both the bings and a wide range of surrounding habitats, *Holcus lanatus*, *Centaurea nigra* and *Plantago lanceolata,* are all perennials. *H. lanatus* and *C. nigra* were both demonstrated to be extensive producers of seed from this study (Table 5.3). *H. lanatus* produces large numbers of seeds in each flower-head and *C. nigra* produces large numbers of flowers on each plant. *P. lanceolata* is less productive than the other two species and this is reflected in the lower density of plants but was shown to have a high success rate in the germination and seedbank trials. High seed production and germination success, combined with vegetative regeneration and longevity, leads to the high density of plants, in all three species, recorded on the bings. The same factors are probably responsible for the general abundance of these species throughout Britain.

The apparent absence of *C. nigra* in the seedbank of all three sites in this study was unpredicted. The large number of seeds produced in the field and high germination rates in the glass house trials suggest that there should be a noticeable presence of this species in the seedbank. Seeds tend to be retained in the head of this species and are shed at irregular times throughout autumn and winter. The seeds are known to be heavily predated both by insects, in the capitula, and small rodents, on the ground (Grime *et al*., 1988) although Fenner *et al*. (2002) recorded a much lower percentage of insect infestation in this species compared with the two Asteraceae species (12.7 ± 4.0). *C. nigra* may rely more heavily on longevity and permanence of individuals on the bings than the other species.

Individuals of *P. lanceolata* were recorded in the field at almost twice the published normal maximum height of 500 mm (Stace, 1997). Height was not correlated with any of the measured environmental factors but particularly tall individuals with additional leaf rosettes on the scape and multiheaded forms were recorded at Oakbank. There is some debate as to whether this unorthodox development is genetic or caused by galls but it is not normally associated with increased height (Cragg-Barber, 1999). Plants with this aberration were consistently found in close proximity to phyllodic clover and the two species could form the basis of an innovative, teratological investigation.

5.7.6 General observations

This chapter set out to address a series of questions that would help to explain some of the variance in species distribution on the shale bings that had not been associated with physical or chemical environmental gradients and to determine the roles of inherent plasticity, seed production, seed persistence and seedbank in the observed distribution of nine common weed species on shale bings.

In all nine species there is a massive discrepancy between the potential for seedlings, measured as germinable seed produced per m^2 and the density of plants recorded in the field. This suggests high seed and seedling predation or mortality through other causes. Pre-dispersal seed predation by insects has been shown by Reader (1993) to be responsible for up to 90% of seed loss and a similar percentage of seeds are lost to rodents post-dispersal although these percentages vary considerably between species.

The unsuccessful seed rain trials and resultant lack of data from the seed traps was unfortunate especially in the light of the apparent inconsistencies in seed losses between the stages of seed development in all species. A range of alternative seed trap designs were considered before finally adopting the seed traps described in section 5. Many of the commonly used seed trap designs have been developed for use in forestry and are large and noticeable structures that are impractical for use in open habitats, like the shale bings, that are well-used by members of the public. In addition there are intentional biases in the designs to favour the trapping of tree seeds: small and wind-blown seeds when using sticky traps and bird dispersed seeds

when using bucket traps (Kollmann and Goetze, 1998). The design adopted in this study was an adaptation of Schott's (1995) seed trap and was selected because it was low lying and inconspicuous, inexpensive and minimised predation (allegedly).

The collection of quantitative data on the composition of seed rain on the shale bings should be attempted in future studies when a new, crow-proof, seed trap design has been developed and tested in the field. In addition to providing results that could be compared with seed trap trials from literature and objective data to support (or refute) the published seed loss from predation, future analysis of seed rain collections could be used to differentiate between locally produced seed from incomers using genetic markers. For example information on the provenance of the *Senecio viscosus* plants recorded on the bings could provide evidence that would establish if these very small plants are examples of the native dwarf variety alluded to by Preston *et al*. (2002).

There has been a strong inference in this chapter that successfully germinated seedlings will ultimately become mature plants. Seedling mortality on the shale bings has not been considered in the explanation of loss of individuals between seed production and numbers of mature plants recorded in the field and is probably extremely high. Exposure and desiccation are only two obstacles that seedlings have to survive. Abiotic factors like these are likely to have a greater influence on the distribution and abundance of colonising seeds and spores than biotic factors. Jumponnen *et al*., (1999) cite microsites with concave surfaces, coarse surface substrate and those in the vicinity of large rocks as most likely to entrap available seed but the microsites where seeds are trapped must be conducive to germination. The sparse cover of vegetation on many parts of the bings makes them ideal sites for further investigation into the effects of microclimate on seedling survival by comparing surfaces that have and have not been invaded.

The weak correlations between the measured factors of the physical and chemical environment and the variations in height and seed development of the nine species warrant further investigation. It was expected that data collected from these nine common species would indicate that morphology and productivity follow a pattern, or patterns, relating to nutrient availability or physical environment that reflects

stages of soil development and would therefore go some way towards explaining the variance in distribution of vegetation generally. The nine species have demonstrated different relationships between abundance, plant height, number of flower-heads produced and number of seeds produced and the results of the multivariate analyses suggest that the measured environmental factors are influencing the physiology of each species independently.

It must be stressed, however, that although these relationships can not be extrapolated to produce a model for all species in all habitats the analyses of the data demonstrated that the productivity and dispersal mechanisms in each of the nine species studied are significant factors in their distribution (Section 5.6.1) and that almost half the variance in distribution patterns is explained by a combination of physical and chemical environmental factors (Section 5.6.2).

5.7.7 Conclusions

There were almost unlimited interesting issues that could be developed from the data already collected from this study and several of these have been proposed for future investigations throughout this and the preceding chapters. Many factors have not been taken into account in the investigation of the nine species. Above ground morphology expressed as longevity, vegetative spread and persistence are additional properties of perennial plant species that are important in determining relative abundance (Mitchley and Grubb, 1986) and the plasticity within these species is expected to reflect nutrient availability (Grubb, 1987). Annual ruderals may reproduce quickly enough and with sufficient genetic variation to adapt to the non-optimal conditions found on the bing surface within a few growing seasons. The effects of competition, both above and below ground, may have a greater influence on the distribution of species than expected from the sparse vegetation and large expanses of bare substrate recorded on the bings. Unfortunately there were insufficient resources within the remit of this PhD to expand these hypotheses and questions.

The studies have determined that availability of seeds and methods of dispersal are a major determining factor in the establishment of vegetation on the bings. The field

measurements, trials and analysis have established that the height of individual plants within the nine selected species and also the numbers of flowers heads and seeds produced are directly affected by local site conditions. Almost half of all of the variance in physiology in the nine species can be explained by the variables of physical and chemical environment that were measured in this study. The phenotypic plasticity and mechanisms of productivity in each these species respond to different elements of the environment making it impossible to create a generic model of all species. However the study has determined the range of variance that can be expected in plant height, flower and seed production and the percentage of variance in each of these that is explained by one or more of five physical and six chemical environmental variables for each of nine common plant species selected from 211 species growing on the oil-shale bings.

If every one of the species recorded in the baseline survey was researched in the same detail as the nine selected species the combined information collected would be unlikely to provide any ecologically significant additional information on vegetation dynamics. Instead of delving more deeply into the minutiae of individual species the decision was made to take a more holistic approach by modelling the mechanisms and processes of the successional progression on the bings that have been discussed in this and earlier chapters and comparing them with other successional sites, both natural and man-made. A model based on the findings of this thesis would also establish whether the colonisation of the bings follows the accepted models of succession from literature.